

AMENDMENTS TO THE CLAIMS

Please amend claims 1, 22, and 38 as set forth below. Claims 2-8, 14, 17-21, and 33 were previously canceled, without prejudice or disclaimer.

The current listing of claims replaces all prior listings.

1. (Currently Amended) A human embryoid body derived (EBD) cell characterized by: forming disaggregated single cells upon dissociation from embryoid bodies (EB) and adhesion to a defined substrate lacking a feeder layer; having the ability to be maintained in culture on the defined substrate in the absence of a feeder layer; and lacking telomerase activity ~~human embryoid body derived (EBD) cells derived from a human embryoid body, wherein the embryoid body is derived from embryonic germ cells, which are further derived from primordial germ cells, and wherein the EBD cells proliferate in culture upon enzymatic disaggregation from the embryoid body.~~

2.-8. (Canceled)

9. (Previously Presented). The EBD cell of claim 1, wherein under suitable cell culture conditions the EBD cells proliferate for at least thirty population doublings without being immortal under said conditions.

10. (Previously Presented) The EBD cell of claim 9, wherein the EBD cells proliferate for at least sixty population doublings.

11. (Previously Presented) The EBD cell of claim 1, wherein the EBD cells proliferate under suitable cell culture conditions that are nonpermissive for proliferation of human embryonic germ cells.

12. (Previously Presented) The EBD cell of claim 1, wherein the EBD cells proliferate under suitable cell culture conditions lacking leukemia inhibitory factor, a fibroblast feeder layer, or both.

13. (Previously Presented) The EBD cell of claim 1, wherein the EBD cells are transfectable with a retrovirus or a lentivirus or both.

14. (Canceled)

15. (Previously Presented) The EBD cell of claim 9, wherein the EBD cells are clonal.

16. (Previously Presented). The culture of claim 15, wherein the EBD cells are clonally derived from a single EBD cell.

17.-21. (Canceled)

22. (Currently Amended) A method of obtaining a human embryoid body derived (EBD) cell comprising:

- (a) culturing primordial germ cells under conditions that are suitable for formation of a solid or cystic embryoid body having a 3-dimensional morphology;
- (b) disaggregating the solid or cystic embryoid body under suitable enzymatic conditions to provide a constituent cell or embryoid body derived (EBD) cell; and
- (c) culturing the EBD cell under conditions suitable to produce a population of proliferating EBD cells

wherein the cell is characterized as forming non-aggregated single cells upon dissociation from embryoid bodies (EB) and adhesion to a defined substrate lacking a feeder layer; having the ability to be maintained in culture on the defined substrate in the absence of a feeder layer; and lacking telomerase activity.

23. (Previously Presented) The method of claim 22 comprising selecting a single EBD cell from the EBD cells and culturing the single EBD cell to produce a clonal population of cells.

24. (Previously Presented) The method of claim 22 comprising culturing the EBD cell in a media comprising human basic fibroblast growth factor.

25. (Previously Presented) The method of claim 24 comprising culturing the EBD cell in a media selected from the group consisting of RPMI 1640 supplemented with 15% FCS and media consisting essentially of hEGF, hydrocortisone, gentamicin, amphotericin-B, fetal bovine serum, VEGF, hFGF-2, heparin, recombinant human IGF-1 and ascorbic acid.

26. (Previously Presented) The method of claim 25 comprising culturing the EBD cell in a media consisting essentially of hEGF, hydrocortisone, gentamicin, amphotericin-B, fetal bovine serum, VEGF, hFGF-2, heparin, recombinant human IGF-1 and ascorbic acid.

27. (Previously Presented) The method of claim 22 comprising culturing the EBD cell on a matrix.

28. (Previously Presented) The method of claim 27 comprising culturing the EBD cell on a matrix that is selected from the group consisting of collagen I, human extracellular matrix extract, and tissue culture-treated plastic.

29. (Previously Presented) The method of claim 28 comprising culturing the EBD cell on a matrix selected from the group consisting of collagen I and human extracellular matrix.

30. (Previously Presented) The method of claim 22 comprising culturing the EBD cell on a media that is not permissive for proliferation of the EG cells.

31. (Previously Presented) The method of claim 30 comprising culturing the EBD cell on a media lacking leukemia inhibitory factor, a fibroblast feeder layer, or both.

32. (Previously Presented) The method of claim 22 comprising culturing the population of proliferating EBD cells for at least 30 population doublings.

33. (Canceled)

34. (Previously Presented) The EBD cell of claim 1, wherein the enzyme includes collagenase, dispase, or both.

35. (Previously Presented) The method of obtaining a human EBD cell of claim 22, wherein the enzyme includes collagenase, dispase, or both.

36. (Previously Presented) The method of claim 22, further comprising expanding the proliferating cells on a matrix.

37. (Previously Presented) The method of claim 36, wherein the matrix is selected from the group consisting of collagen I, human extracellular matrix extract, and tissue culture-treated plastic.

38. (Currently Amended) A method of obtaining a human embryoid derived (EBD) cell comprising:

- (a) culturing primordial germ cells under conditions that are suitable for formation of a solid or cystic embryoid body having a 3-dimensional morphology;
- (b) disaggregating the solid or cystic embryoid body under suitable enzymatic conditions to provide a constituent cell or embryoid derived (EBD) cell; and
- (c) expanding the EBD cell under conditions suitable to produce a population of proliferating EBD cells, wherein the EBD cells proliferate on the matrix, and wherein the

In re Application of:
Shambrott and Gearhart
Application No.: 10/767,421
Filing Date: January 22, 2001
Page 6

PATENT
Attorney Docket No. JHU1750-1

matrix is selected from the group consisting of collagen I, human extracellular matrix extract, and tissue culture-treated plastic,
wherein the EBD cell forms non-aggregated single cells upon dissociation from embryoid bodies, whereby the EBD cell will adhere to a defined substrate lacking a feeder layer, and whereby the EBD cell lacks telomerase activity.